Clean Copy of Amended Title

MP121, A GROWTH/ DIFFERENTIATION FACTOR OF THE TGF-β FAMILY

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C1 BRIEF DESCRIPTION OF THE FIGURES

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Figure 3 shows a diagram of a Western blot using chicken antibodies against human MP121. Lane 1 shows *E. coli* cells transformed with pBP4MP121His under reducing conditions (1% β-mercaptoethanol. Lane 2 shows cell culture supernatant of NIH-3T3 cells after infection with recombinant viruses (with inserted MP121 cDNA) under reducing conditions (1% β-mercaptoethanol). Lane 3 shows cell culture supernatant of NIH-3T3 cells after infection with recombinant viruses (with inserted MP121 cDNA) under non-reducing conditions. Lane M shows prestained protein molecular weight markers having the stated apparent molecular weights (Gibco BRL #26041-020).

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Figure 4 shows the expression of MP121 compared to activin β_A and β_B in various mouse tissues and is an autoradiogram after gel analysis of an RNase protection assay using specific probes against activin βA (βA), activin βB (βB), MP121 and against GAPDH for the control. Total RNA was tested which has been isolated from various mouse tissues (Lane 1: brain; Lane 2: heart, Lane 3: kidney, Lane 4: liver, Lane 5: lung, Lane 6: muscle, Lane 9: ovary, Lane 10: spleen, Lane 11: testes) from embryonic stem cells (Lane 12: CJ7) and from yeast (Lane 13) as a control. No RNA was used in Lane 14 as a control. The unprotected antisense RNA probes used for the hybridization

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are applied in lanes 8 and 15 and the expected fragment size is indicated in brackets in the right margin. The bands of the protected fragments are labeled in the left margin. PBR322 restricted with Map 1 (Biolabs #303) and end-labeled with γ-32p-ATP (Amersham) was used as the marker (Lane 7).

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Figure 5 shows a positive influence on the survival of dopaminergic neurons by treatment with partially purified MP121. The number of TH-immunoreactive dopaminergic neurons surviving after isolation from the mesencephalon of rat embryos (E14) after 8 days culture is shown. The effect of 20 ng/ml partially purified MP121 was tested compared to the equivalent amount of partially purified control supernatant (wt) as well as untreated neurons (control: medium containing 0.3% acetonitrile). The mean ± SEM from a triple determination is shown.

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Figure 6 shows a Western blot using rabbit antibodies against human MP121. Lane 1 shows cell culture supernatant of HepG2 cells after infection with recombinant viruses (with inserted MP121 cDNA) under non-reducing conditions. Lane 2 shows cell culture supernatant of HepG2 cells after infection with wildtype viruses under non-reducing conditions. Lane 3 shows prestained protein molecular weight markers having apparent molecular weights of 15.5, 18.2, 27.8, 43.8 and 71.5 kD (Gibco BRL #26041-020), indicated schematically. Lane 4 shows cell culture supernatant of HepG2 cells after infection with recombinant viruses (with inserted MP121 cDNA) under reducing conditions. Lane 5 shows cell culture supernatant of HepG2 cells after infection with wildtype viruses under reducing conditions.

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C6 Figure 7 shows the stimulation of nerve fibre outgrowth from the embryonic retina by treatment with partially purified PM121. Dark field microscopy of living cultures shows nerve fibre outgrowth from explanted chicken retina after 4 days in culture in the presence of 5 ng/ml partially purified MP121.

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C7 Figure 8 shows that various concentrations of partially purified MP121 can inhibit EGF induced DNA synthesis in hepatocytes.

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C8 Figure 9 shows the influence of various concentrations of partially purified MP121 on erythroid differentiation measured by the percentage of dianisidine positive cells.